Fish Tissue Metal Concentrations

Data were analyzed on metal concentrations (Aluminum, Arsenic, Cadmium, Copper, Lead, Mercury, Manganese, and Selenium) in fish that were collected in August 2015 and March 2016 by the New Mexico Department of Game and Fishes using raft electrofishing. Analysis was done using a generalized linear model (GLM) routine in R. Categorical factors examined included site, sampling date, fish species and type of tissue. Body length was included as a continuous independent variable. Blue-shaded cells in Table 1 indicate the "reference" level of each categorical factor. Model coefficients estimated for each level of a categorical factor represent the difference between that level and the reference level, for which no model coefficient is estimated (it's impact on the response variable is wrapped into the model intercept).

Table 1. Independent variables examined in the generalized linear model for metal concentrations in fish tissues in the lower Animas and San Juan rivers collected in 2015 and 2016.

Site	Tissue	Collection Date	Species	Fish Length (mm)
Animas River RK 148	Muscle	August 2015	Bluehead Sucker	
Animas RK 163	Liver	March 2016	Brown Trout	
San Juan RK 196			Flannelmouth Sucker	
San Juan RK 214			Speckled Dace	
San Juan Reference				

The reference site on the San Juan River is location several kilometers upstream of the San Juan/Animas River confluence in Farmington, NM. Results of the GLM analysis are given in Table 2, which provides the estimated model coefficients (in mg/kg) and denotes the significance of each term in the model. Blue-shaded cells in the table indicate significantly lower metal concentrations that the reference level, and yellow-shaded cells indicate significantly higher metal concentrations than the reference level. For example, the analysis found that the aluminum concentration in fish collected in March 2016 was, on average, 57 mg/kg less than fish collected in August of 2015. Brown Trout had, on average, 80 mg/kg higher Copper concentrations than Bluehead Sucker, etc.

Table 2. Generalized linear model results for metal concentrations in fish. Cell values are estimated model coefficients in mg/kg. Blue-shaded cells indicate a statistically significant model coefficient that is less than the reference level of the factor (given in parentheses in the gray "Factor" heading). Yellow-shaded cells indicate a statistically significant model coefficient that is greater than the reference level of the factor. For the continuous variate Total Length, blue-shaded cells indicate a significant negative relationship between body length and metal concentration, while yellow-shaded cells represents a significant positive relationship between those variables.

Factor	Aluminum	Arsenic	Cadmium	Copper	Lead	Mercury	Manganese	Selenium
Location (SJ Reference)								
River KM 148	54	0.032	-4.9	16.4	0.73	-0.093	15	0.62
River KM 163	105	0.029	-2.6	7.7	0.23	-0.088	14.2	0.23
River KM 196	15	0.0056	0.29	3.2	0.00054	-0.071	1.1	0.09
River KM 214	27	0.05	2.5	3.5	0.15	-0.081	3.9	0.2
Collection Date (August 2015)								
March 2016	-57	-0.024	-6.8	-0.85	-0.52	0.0045	-10.7	-0.77
Tissue (Liver)								
Muscle	-88	-0.168	-6.7	-39.3	-0.75	0.011	-21.9	-3.4
Species (Bluehead Sucker)								
Brown Trout	-84	-0.213	3.7	80	-0.86	-0.0018	-22.7	6.8
Flannelmouth Sucker	-24	-0.15	5.3	9	-0.19	0.0071	-7.6	0.41
Speckled Dace	-156	-0.21	12.5	27.8	-1.1	0.062	-26.3	3.1
Fish Size								
Total Length (mm)	-0.49	-0.00047	0.044	0.0365	-0.0035	0.00016	-0.087	0.0042

Site. Significantly higher Mercury concentrations were found in fish collected at the San Juan reference site relative to the other four sites (i.e., the entire Mercury column within the Location factor is blue). Fish collected at the two Animas River sites had significantly higher Manganese concentrations than the San Juan reference site. The two San Juan sampling sites below the confluence with the Animas River showed no difference in metal concentrations relative to the San Juan reference site, outside of having lower Mercury concentrations. The fish collected on the Animas River at kilometer 163 (near Aztec, NM) had higher aluminum concentrations than fish collected at any other site.

Collection Date. For most metals tested, March 2016 metal concentrations were significantly less than August 2015 concentrations. Concentrations in fish are responsive concentrations in the water, so this result is not a surprise due to the higher water concentrations of these metals in August 2015, just after the GKM release event, relative to March 2016.

Tissue. For all metals except Mercury, significantly higher metal concentrations were found in liver samples relative to muscle tissue. Most metals bind readily to compounds found in the liver (metallothioneins), while mercury also binds to thiols/sulfhydryls commonly found in muscle tissue. This result is well supported in scientific literature.

Species: A mixture of results were seen for this factor. Bluehead sucker had significantly higher concentrations of Aluminum, Arsenic, Lead and Manganese, but significantly lower concentrations of Cadmium, Copper, and Selenium. This could be attributed to the ecological niche (habitat preferences, diet, trophic status, etc.) of this species compared to the other species. Whereas the Brown Trout and Speckled Dace showed significant statistical differences in metal concentrations across nearly all the metals relative to the Bluehead sucker, the Flannelmouth sucker had only two significant differences: lower Arsenic and higher Cadmium. This is not surprising given these two sucker species (sharing a taxonomic family) are more ecologically similar than the other two species that were sampled.

Fish Size. Metal concentrations declined with fish size for four metals (Aluminum, Arsenic, Lead and Manganese), increased with fish size for two metals (Cadmium and Mercury), and showed no relationship to size for two metals (Copper and Selenium). We propose that these patterns can be explained by examining the varying propensity of each metal to bind to body tissues and bioaccumulate through time (Table 3). A foundation of this explanation is that the % water composition (by weight) within any fish species tends to decline with increasing size. For this exercise, we assume that the concentration of metal in the water inside the fish is at equilibrium to the concentration of metal in the water surrounding the fish, and that concentration is a constant 1 mg/l.

Panel A of Table 3 shows a calculation of whole body concentration for a hypothetical metal that doesn't readily bind to tissues (note the low tissue concentration relative to water concentration), and does not bioaccumulate (no increase in tissue concentration with size). This hypothetical fish species ranges in weight from 10-100 grams, and over this span its % water weight declines from 85% to 67% (adult fish, depending on species, generally are 60-90% water by weight). As the last column of panel A shows, these parameters lead to a situation where metal concentrations decline with increasing fish size.

Panel B shows a scenario where the metal binds more strongly to tissues (note the higher tissue concentrations than in Panel A at the same water concentration), and bioaccumulates to some extent (an increase of 0.36 mg/kg in tissue metal concentration over the size range of the species). In this instance, there would appear to be virtually no relationship between metal concentration and body size.

Finally, in Panel C we show a metal that exhibits higher bioaccumulation in tissues over time. Under this scenario, metal concentrations increase with increasing fish size.

In reality, there is a great deal of variability in how strongly various metals bind/bioaccumulate in organic tissues, the external water concentrations that fish are exposed to on a daily basis, the range in body size within a fish species, and the range in % water weight within that size range. This exercise simply shows that one can easily generate scenarios that produce a range of outcomes (declines/increases/no changes in metal concentrations with fish size) by varying these parameters, so it should be no surprise to see a variety of outcomes within any particular dataset representing a range of different metals and fish species. It would be interesting to see if these relationships are consistent for any metals across datasets, but because of variability in the species sampled and ambient water concentrations, it would not be entirely surprising to find discrepancies.

Table 3. Relationships between fish size and whole-body metal concentrations under varying hypothetical scenarios of metal binding and bioaccumulation within body tissues.

PANEL A

Weight (g)	% Water	Body Mass Water (mg)	Body Mass Tissue (mg)	Water Conc (mg/l)	Tissue Conc (mg/kg)	Metal Mass Water (mg)	Metal Mass Tissue (mg)	Total Body Conc (mg/kg)
10	85	8500	1500	1	0.05	8500	75.0	0.86
20	83	16600	3400	1	0.05	16600	170.0	0.84
30	81	24300	5700	1	0.05	24300	285.0	0.82
40	79	31600	8400	1	0.05	31600	420.0	0.80
50	77	38500	11500	1	0.05	38500	575.0	0.78
60	75	45000	15000	1	0.05	45000	750.0	0.76
70	73	51100	18900	1	0.05	51100	945.0	0.74
80	71	56800	23200	1	0.05	56800	1160.0	0.72
90	69	62100	27900	1	0.05	62100	1395.0	0.71
100	67	67000	33000	1	0.05	67000	1650.0	0.69

PANEL B

Weight (g)	% Water	Body Mass Water (mg)	Body Mass Tissue (mg)	Water Conc (mg/l)	Tissue Conc (mg/kg)	Metal Mass Water (mg)	Metal Mass Tissue (mg)	Total Body Conc (mg/kg)
10	85	8500	1500	1	0.30	8500	450	0.90
20	83	16600	3400	1	0.34	16600	1156	0.89
30	81	24300	5700	1	0.38	24300	2166	0.88
40	79	31600	8400	1	0.42	31600	3528	0.88
50	77	38500	11500	1	0.46	38500	5290	0.88
60	75	45000	15000	1	0.50	45000	7500	0.88
70	73	51100	18900	1	0.54	51100	10206	0.88
80	71	56800	23200	1	0.58	56800	13456	0.88
90	69	62100	27900	1	0.62	62100	17298	0.88
100	67	67000	33000	1	0.66	67000	21780	0.89

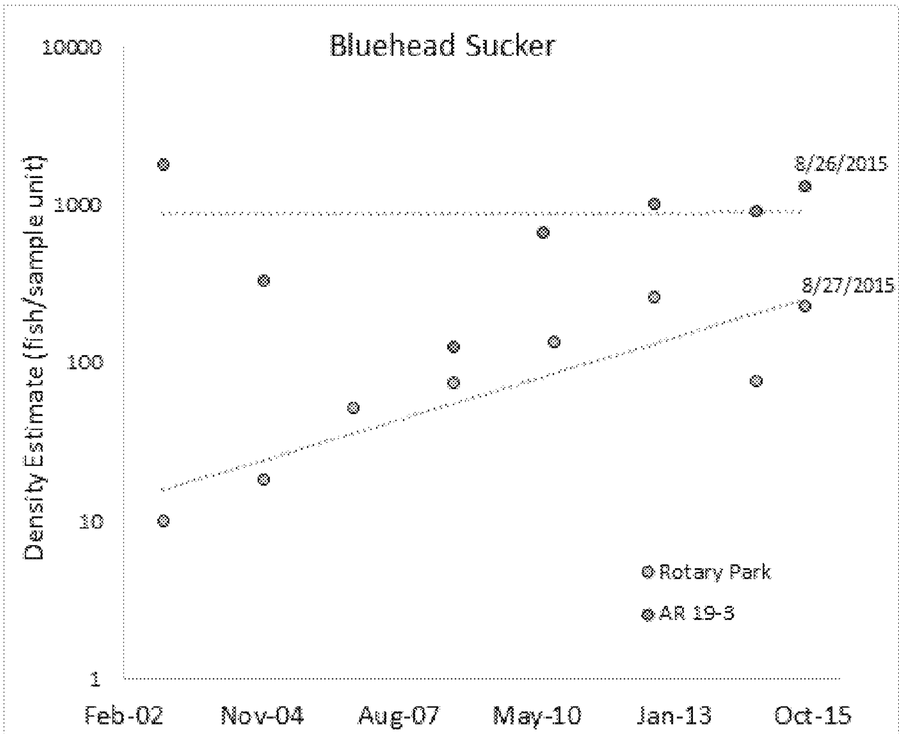
PANEL C

Weight (g)	% Water	Body Mass Water (mg)	Body Mass Tissue (mg)	Water Conc (mg/l)	Tissue Conc (mg/kg)	Metal Mass Water (mg)	Metal Mass Tissue (mg)	Total Body Conc (mg/kg)
10	85	8500	1500	1	0.3	8500	450	0.90
20	83	16600	3400	1	0.5	16600	1700	0.92
30	81	24300	5700	1	0.7	24300	3990	0.94
40	79	31600	8400	1	0.9	31600	7560	0.98
50	77	38500	11500	1	1.1	38500	12650	1.02
60	75	45000	15000	1	1.3	45000	19500	1.08
70	73	51100	18900	1	1.5	51100	28350	1.14
80	71	56800	23200	1	1.7	56800	39440	1.20
90	69	62100	27900	1	1.9	62100	53010	1.28
100	67	67000	33000	1	2.1	67000	69300	1.36

Population Density Estimates

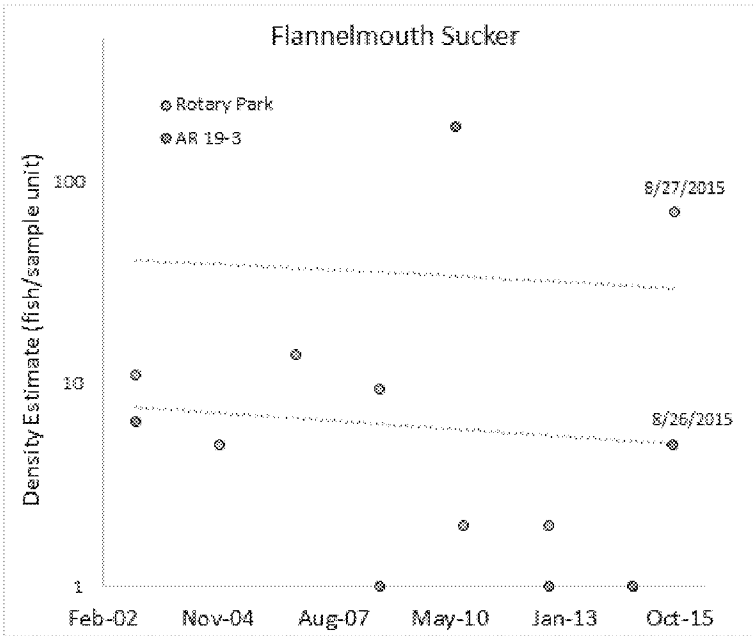
We obtained a dataset of fish density estimates (# of fish per sampling unit) made by the Colorado Parks and Wildlife (CPW) agency based on multiple-pass electrofishing and mark-recapture studies at two primary locations on the Animas River near Durango, CO: Rotary Park (RK 95) and AR 19-3 (RK 102). Although these data included samples taken as early as August 1912, we focused on estimates made every few years during the period 2002-2015. In addition, we only examined those species for which population estimates were consistently made at both locations across that time span: Bluehead Sucker, Flannelmouth Sucker, White Sucker, Mottled Sculpin, Brown Trout and Rainbow Trout. We plot density estimates through time and look for visual evidence that the most recent estimates made in late-August 2015 indicate a significant population decline, possibly due to the GKM release event.

Bluehead Sucker



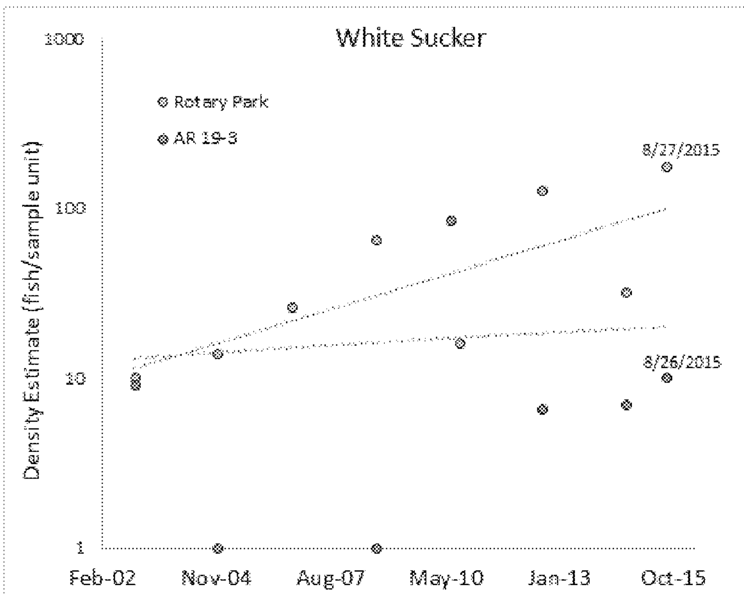
Densities of Bluehead Sucker at AR 19-3 (orange dots) show a dip in the middle of the sampling period and a flat to slightly increasing trend since 2010, while a general increasing trend is evident at Rotary Park (blue dots). The two areas have more similar densities lately as compared to earlier in the sample period, when the population looked much denser at AR 19-3. The August 2015 samples at both sites are very near to being the largest densities at their respective sites across the sampling period.

Flannelmouth Sucker



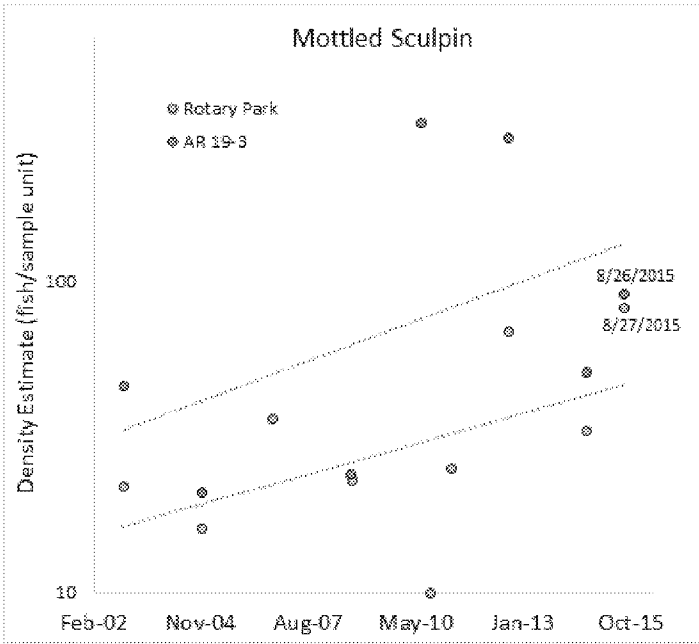
Other than a large aberrant value in 2010, densities of Flannelmouth Sucker at AR 19-3 were lower than densities at Rotary Park. The 2015 estimate at AR 19-3 shows a large increase above the two previous low estimates in 2012 and 2014. The 2015 estimate at Rotary Park is by far the greatest of this sampling period at that site.

White Sucker



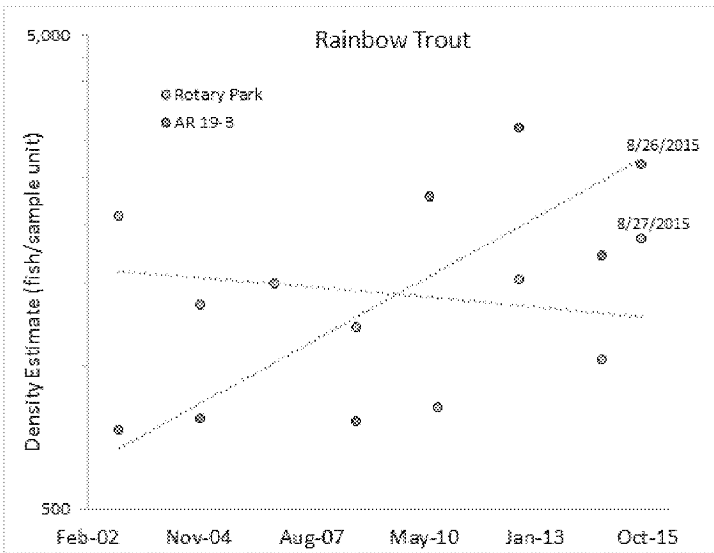
As with Flannelmouth Sucker, densities at Rotary Park tended to be higher than at AR 19-3 for White Sucker. The 2015 measure at Rotary Park was the highest of the series, and fits into the increasing temporal trend at this site. The 2015 estimate at AR 19-3 is slightly higher than the two most recent estimates in 2012 and 2014, but not close to the large estimate made in 2010.

Mottled Sculpin



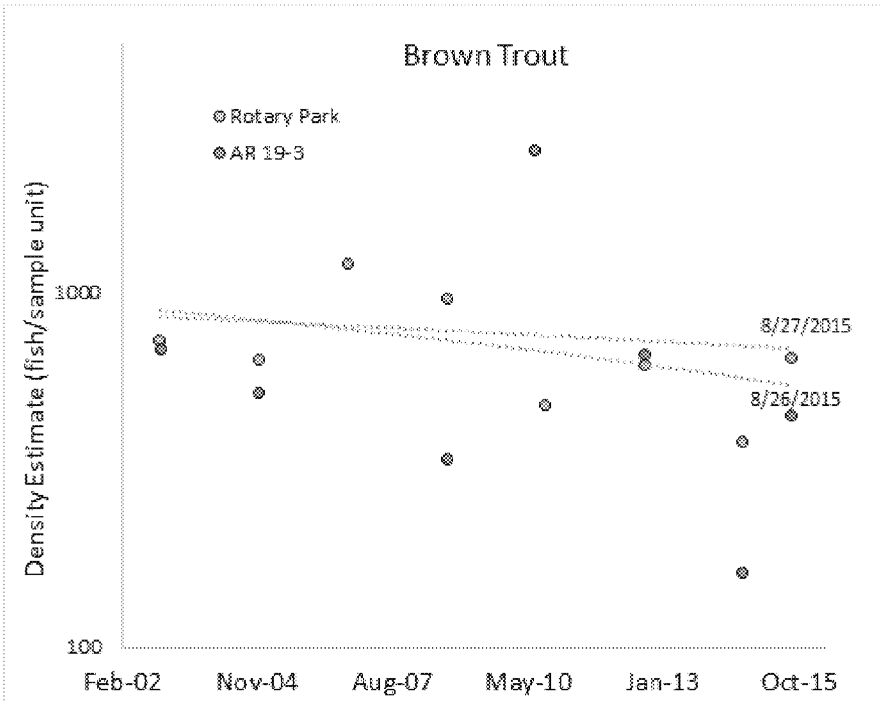
Mottled Sculpin densities are generally higher at AR 19-3 relative to Rotary Park, but the 2015 estimates are very similar to one another. The 2015 estimate at Rotary Park is the largest of the series for that site; the 2015 estimate at AR 19-3 is not as large as two estimates made in 2010 and 2012, but greater than the estimate made in 2014.

Rainbow Trout



Rainbow Trout exhibit a shift midway through the sampling period, with greater densities at Rotary Park prior to 2008, and greater densities at AR 19-3 after that time. Both 2015 estimates are nearly (but not quite) the largest densities seen at their respective sites across the sampling period.

Brown Trout



Densities of Brown Trout are generally greater at Rotary Park than AR 19-3 (with 2010 being a very large exception). The temporal trend at both sites is slightly decreasing, but the 2015 estimates at both sites show a large increase compared to the 2014 estimates.

Overall Conclusions of Density Data

We see no evidence of a population crash in 2015 for any of the six species we examined at either of these sites, either through direct mortality from the GKM release event, or emigration from these areas afterwards. However, the lack of evidence for acute, short term population responses to the GKM release does not prove longer term impacts are negligible. The event may have been more impactful on larval and juvenile fish not sampled in these surveys, or on the eggs of species that spawn later in the fall and winter. If so, there could be a reduction in year-class strength for some of the naturally-reproducing fish species inhabiting the Animas River, and reductions in adult populations would only become evident some years later as the affected cohorts mature. Of course, for those trout species (Rainbow and Brown) stocked by the state of Colorado, population levels are determined by stocking rates, not natural spawning events.